#### LIGHT RAY TRACING THROUGH A LEAF CROSS SECTION

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#### Abstract

A light ray, incident at about 5° to the normal, is geometrically plotted through the drawing of the cross section of a soybean leaf using Fresnel's Equations and Snell's Law.

The optical mediums of the leaf considered for ray tracing are: air, cell sap, chloroplast and cell wall. The above ray is also drawn through the same leaf cross section considering cell wall and air as the only optical mediums. The values of the reflection and transmission found from ray tracing agree closely with the experimental results obtained using a Beckman DK-2A Spectroreflectometer.

#### I. Introduction

Willstatter and Stoll ( $\underline{W-S}$ ) in 1918, proposed a theory to explain reflectance from a leaf on the basis of critical reflection of visible light at spongy mesophyll cell wall - air interfaces. According to several authors (i.e., Gates et al.<sup>2</sup> and Gausman et al.<sup>3</sup>) their experimental results on reflectance

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from leaves seem to have supported the W-S theory. Sinclair et al. 4 gave an excellent review of the reflectance and transmittance from the leaves. They critically examined the commonly accepted W-S theory and proposed a modification, termed the "diffuse reflectance hypothesis," which is based on diffusing reflecting qualities of cell walls oriented at near perpendicular angles. 4 They pointed out that the microfibril structure of the cell wall presumably induces the scattering necessary to have diffuse reflectance. They presented experimental results on both the reflectance and transmittance from various species of leaves for both the visible (0.50 to 0.72 µm) and the reflective infrared (0.72 to 1.3 µm) wavelengths, which could not be satisfactorily explained by the W-S theory, but which they felt could be accounted for on the basis of their hypothesis.

Myers and Allen<sup>5</sup> explained the K-M (Kubelka - Munk) scattering coefficient (of diffuse reflectance) for a typical leaf by Fresnel reflections at normal incidence from 35 interfaces along the mean optical path through the leaf. Gausman et al.<sup>6</sup> noted that if oblique reflections are considered, fewer interfaces account for the results. Knipling<sup>7</sup> emphasized that the air spaces within the palisade parenchyma layer of a leaf mesophyll may be more important in scattering light than air spaces in the spongy parenchyma layer. Allen et al.<sup>8</sup> have proposed that the complex structure of the leaf can be simulated

by a pile of transparent plates with Perfectly diffusing surfaces. Birth<sup>9</sup> has given an excellent critical review of existing concepts on the reflectance from a leaf. He pointed out that the work of Sinclair<sup>4</sup> is enlightening in that the diffuse character of light in the leaf is shown to start at the initial interface. Recently, Kumar<sup>10</sup> has reviewed much literature pertaining to reflection from leaves.

The purpose of this investigation is to compare the reflectance of a typical leaf found by tracing the ray of light through the leaf with the experimentally determined reflectance values of the same leaf. In addition, the authors would like to investigate if considering only cell wall and air as the optical mediums in ray tracing leads to good predictions of experimentally determined reflectance of the leaf; and if other optical mediums — cell sap and chloroplasts — should also be included in the ray tracing for significantly better prediction of the reflectance. Furthermore, the authors would like to create a more realistic illustration to show the pathway of a light ray through the leaf than shown by Willstatter and Stoll. 1

## II. Cross Section of the Soybean Leaf

The cross section of the soybean leaf was taken from Sinclair's thesis. 11 This cross section had been obtained by Sinclair by microtome cross-sectioning and a microscopic slide

was prepared using the techniques outlined by Jensen. 12 This cross section was enlarged. An artist, well familiar with the cross section of leaves, drew the above mentioned cross section on a plain paper showing explicitly the cell walls, cell sap and chloroplasts, a part of which is shown in Figure 1. The cross section of Figure 1 was enlarged in order to do ray tracing conveniently and accurately.

#### III. Reflectance From a Leaf

- A. Proposed Leaf Reflectance Model. The following assumptions are made in the reflectance model of a leaf:
  - 1. The leaf is assumed to consist of homogeneous and isotropic media -- cell wall, chloroplasts, cell sap and air. This assumption is made for mathematical simplicity so that Fresnel's Equations can be applied at each interface.
  - 2. Geometrical Optics is assumed to be valid for the media of the leaf mentioned above. This is not quite valid for chloroplasts (typical dimensions 5  $\mu$ m to 8  $\mu$ m in diameter and about 1  $\mu$ m in width<sup>2</sup>) where diffraction is likely to be important.
  - 3. The Rayleigh and Mie scattering by the leaf constituents (of the order of wavelength of light or smaller) is neglected. Gates<sup>2</sup> pointed out that cell dimensions of a leaf

are generally too large for scattering; however, the chloroplasts and grana dimensions are such as to create some scattering (i.e., grana is about 0.5 µm in length and about 0.05 µm in diameter). Scattering coul' also be caused by mitochondria, ribosomes, nuclei, starch grains, and other plastids, etc. It is very hard to take scattering into account because the dimensions, distribution and refractive indices of these particles in the leaf cells are extremely complex and unknown.

4. The absorption of light by the leaf media is neglected. This is quite valid for most leaves in about 0.7 to 1.3 µm wavelength region. Since the leaf media absorb the light in the visible wavelengths, their indices of refraction are complex numbers. The model presented here can also be applied to the visible wavelengths for Fresnel's Equations and Snell's Law are also valid for absorbing media, if one uses the appropriate complex index of refraction. 13 However, at present, it is not possible to do ray tracing in the visible wavelengths since the complex indices of refraction of the leaf constituents in these wavelengths are not yet known. Also, the ray tracing in the visible wavelengths becomes quite involved because the index of refraction, angle of refraction, etc., are complex numbers.

- 5. The two dimensional cross section of a leaf (three dimensional leaf) is used for predicting the reflectance from a leaf.
- B. <u>Basic Equations</u>. Fresnel's Equations, Snell's Law and boundary conditions used for determining reflection and refraction at an interface are given below.

$$n_1 \sin \theta_i = n_2 \sin \theta_r \tag{1}$$

$$R_{\parallel} = \frac{\left[\left(\frac{n_2}{n_1}\right)^2 \cos \theta_i - \left[\left(\frac{n_2}{n_1}\right)^2 - \sin^2 \theta_i\right]^{1/2}\right]}{\left(\frac{n_2}{n_1}\right)^2 \cos \theta_i + \left[\left(\frac{n_2}{n_1}\right)^2 - \sin^2 \theta_i\right]^{1/2}} I_{\parallel} \qquad (2)$$

$$R_{\perp} = \begin{bmatrix} \cos \theta_{i} - \left[ \left( \frac{n_{2}}{n_{1}} \right)^{2} - \sin^{2} \theta_{i} \right] & 1/2 \\ \cos \theta_{i} + \left[ \left( \frac{n_{2}}{n_{1}} \right)^{2} - \sin^{2} \theta_{i} \right] & 1/2 \end{bmatrix}$$

$$I_{\perp} \qquad (3)$$

$$R = \frac{R_{||} + R_{||}}{2} \tag{4}$$

$$\mathbf{I}_{||} = \mathbf{I}_{||} - \mathbf{R}_{||} \tag{5}$$

$$\mathbf{T}_{\perp} = \mathbf{I}_{\perp} - \mathbf{R}_{\perp} \tag{6}$$

$$T = \frac{T_{\parallel} + T_{\perp}}{2} \tag{7}$$

where

n, = refractive index of the first medium

n, = refractive index of the second medium

 $\theta_i$  = angle of incidence

 $\theta_{\perp}$  = angle of refraction

 $R_{ij}$  = reflection parallel to the plane of incidence

 $R_{\parallel}$  = reflection perpendicular to the plane of incidence

R = total reflection

I = incident intensity parallel to the plane of incidence

I<sub>1</sub> = incident intensity perpendicular to the plane of incidence.

 $T_{||}$  = transmission parallel to the plane of incidence

 $T_{\parallel}$  = transmission perpendicular to the plane of incidence

T = total transmission

# C. Indices of Refraction of Leaf Constituents.

The index of refraction of the air spaces in the leaf cells is assumed to be one. The refractive index of a potato cell wall was found to be equal to 1.52 by Birth 14 in the green

wavelengths by Index Matching Technique (i.e., The cell wall was infiltrated with various liquids, mostly oils, having varying refractive indices. The minimum reflectance was noted visually with a medium having a refractive index of 1.52, which was taken to be the best approximation to the refractive index of the potato cell wall.). The value of the index of refraction of the cell wall of the soybean leaf was assumed to be equal to 1.52 for the purpose of ray tracing, as it is expected to be quite close to the refractive index of the potato cell wall. The values of refractive indices for cell sap and chloroplasts were taken from Charney and Brackett 15 to be equal to 1.36 and 1.42, respectively. The values of the index of refraction of the leaf constituents in the 0.7  $\mu m$   $\sim$  1.3  $\mu m$  region are not available because it is quite difficult to measure the refractive indices of the leaf constituents by the Index Matching Technique in the infrared wavelength region as the human eye cannot see in that region. The value of the real part of the index of refraction of water is roughly the same in the near infrared region 16 (i.e.,  $0.7 \mu m \sim 1.3 \mu m$ ) as in the visible wavelength region within .01. Since water is the main constituent of the cell wall, cell sap and chloroplasts, and since none of these absorb light strongly in the 0.7  $\mu m \sim 1.3 \mu m$  region, the refractive indices of these constituents were assumed to be the same in the 0.7  $\mu m$   $\sim$  1.3  $\mu m$ 

region as in the visible wavelength region.

- D. Method of Ray Tracing. The four leaf constituents -cell wall, chloroplasts, cell sap and air -- give rise to the
  following eight optical interfaces in the leaf all of which
  were considered in the ray tracing: 1) air to cell wall,
- 2) cell sap to cell wall, 3) chloroplasts to cell wall,
- 4) cell sap to chloroplasts, 5) chloroplasts to cell sap,
- 6) cell wall to chloroplasts, 7) cell wall to cell sap, and
- 8) cell wall to air.

In ray tracing, a ray of light of intensity  $I_{|||}$  (intensity parallel to the plane of incidence) = 1.000, and  $I_{|||}$  (intensity perpendicular to the plane of incidence) = 1.000 at about 5° to the normal was taken. The angle was taken 5° to the normal, because in the experimental set up with the DK-2A spectroreflectometer the light rays were incident at 5° to the leaf normal. A tangent and a normal were drawn at the interface. The angle of incidence of the ray was measured with a drafting set which can measure angles up to an accuracy of 5 minutes. Knowing the angle of incidence and relative index of refraction at the interface, the values of  $\theta_{r}$ ,  $R_{||}$ ,  $R_{||}$ ,  $T_{||}$ , and  $T_{||}$  were found using equations given in 3B, and the refracted and reflected rays were drawn. Similar procedure was followed at the subsequent interfaces. Each ray was continued until it ended up as reflection

or transmission from the leaf. The rays whose total intensity became less than 0.018 were discontinued to reduce the time and efforts required in ray tracing.

The light ray passed through a total of 253 interfaces out of which total internal reflection took place at 18 cell wall - air interfaces, two cell wall - chloroplast interfaces, and one cell wall - cell sap interface.

Table 1(a) shows the values of the reflected and transmitted intensity of the ray at the first seven interfaces. The pathway of the ray in a part of the leaf cross section, as given by this model, is shown by solid lines in Figure 1. The numbers along the rays represent their total intensity. For simplicity, only the rays whose total intensity is more than 0.018 are shown in the diagram. It can be understood from the Figure 1 that if one takes a number of parallel rays incident on the leaf, each ray will encounter different geometrical internal surfaces and consequently will be reflected and transmitted in different directions. That is how a collimated beam of light incident on the leaf keeps on becoming diffuse slowly as it passes through the leaf. The greater the number of interfaces the light rays encounter in their path, the more diffuse the rays are likely to The pathway of light rays as envisioned by Willstatter and Stoll is shown in Figure 2. It can be seen from Figure 2 that the light rays pass through the epidermis and palisade cells

without any deviation, which is unrealistic. Furthermore,
Willstatter and Stoll did not show the reflection of light at
air - cell wall interfaces, and at cell wall - air interfaces
at angles of incidence less than the critical angle. The
authors would like to emphasize that although cell wall - air
interface causes more deviation of the ray than any other
single interface for a given angle of incidence, and is perhaps
the most important interface for contributing to the reflection
from the leaf, the other interfaces can also contribute
significantly to the reflection from a leaf.

It seems that the reflection of light in the near infrared wavelengths  $(0.7 \sim 1.3 \ \mu\text{m})$  from a typical leaf is likely to be more diffuse than its reflection in the visible wavelengths. This is because the near infrared light rays are likely to pass through many more interfaces of the leaf (because of almost no absorption of light in the near infrared wavelengths) than the corresponding light rays of the visible wavelengths. Also, the transmission from a leaf in the visible as well as near infrared wavelengths is likely to be fairly diffuse because a typical light ray has to pass through a fairly large number of interfaces before it is transmitted. These qualitative conclusions support the experimental results of Breece and Holmes 17 on healthy green soybean and corn leaves.

Ray tracing was also done following the same procedure as the one mentioned above for the same original ray of light (I<sub>||</sub> = 1.000 and I<sub>|</sub> = 1.000) except that only the following two interfaces were considered: 1) air to cell wall and 2) cell wall to air. The light ray passed through a total of 144 interfaces out of which total internal reflection took place at 13 cell wall - air interfaces. Table 1(b) shows the values of the reflected and transmitted intensity of the ray at the first 7 interfaces. The pathway of the ray considering the above two interfaces, in a part of the leaf cross section, is shown in Figure 1 by dotted lines. It can be seen from Figure 1 that the light ray shown by dotted lines follows quite a different path than that shown by solid lines.

## IV. Experimental and Ray Tracing Results

The value of reflection found by Sinclair<sup>11</sup> using a Beckman DK-2A Spectroreflectometer on the same leaf, whose cross section is shown in Figure 1, in the 0.7  $\sim$  1.3  $\mu$ m region, was 47%. Transmission = 100 - 47 = 53% (because absorption of a leaf is almost equal to 0 in the 0.7  $\sim$  1.3  $\mu$ m wavelength region).

# 13 Ray Tracing Results

Note: The values of (reflection + transmission) found were assumed to be 100%.

Reflection (using 8 interfaces = 45.6% mentioned in sec. III D)

Transmission (using 8 interfaces = 54.4% mentioned in sec. III D)

Reflection (using air - cell wall = 30.3% and cell wall - air interfaces)

Transmission (using air - cell wall = 69.7% and cell wall - air interfaces)

Experimental results of Wooley 18 on the soybean leaves strongly support these ray tracing results. Wooley found the reflectance of a soybean leaf in 0.7 ~ 1.3 µm wavelength region to be about 47 percent. But after the soybean leaf was vacuum infiltrated with oil of refractive index 1.48, which essentially eliminated the air to cell wall and cell wall to air interfaces only, its reflectance dropped to about 15 percent. This experiment clearly shows that the reflectance caused by the discontinuities in the indices of refraction of the geometrical surfaces (of the dimensions much larger than the wavelength of light) is significantly more than the reflection caused due to Rayleigh and/or Mie scattering by the particles (of the order of wavelength of light or smaller) inside the leaf cells because the reflectance caused by scattering should essentially remain unchanged after the leaf is vacuum infiltrated with oils of

different refractive indices. Furthermore, it seems to support our conclusion "optical interfaces other than the cell wall to air and air to cell wall can contribute significantly to the reflection from a leaf."

# V. Concluding Remarks

The preliminary conclusions, yet to be confirmed by further ray tracing, and experiments are: considering only cell eall - air and air - cell wall interfaces seems to underestimate the reflection and overestimate the transmission from a leaf significantly in this particular case. Considering all the eight interfaces mentioned in Section III D, ray tracing seems to give results very close to the experimental results. Furthermore, considering only cell wall - air and air - cell wall interfaces is likely to give less diffuse reflectance and transmittance than that given by considering all the eight interfaces. There is some contribution to the reflection from a leaf due to Rayleigh and Mie scattering caused by the particles (of the order of the wavelength of light or smaller) in the leaf cells but the reflection caused by the leaf constituents cell walls, cell sap, chloroplasts, and air, as given by the geometrical optics, is probably more significant than the reflection caused by scattering. Gates<sup>2</sup> pointed out that whatever scattering does exist is probably more of the Mie type than

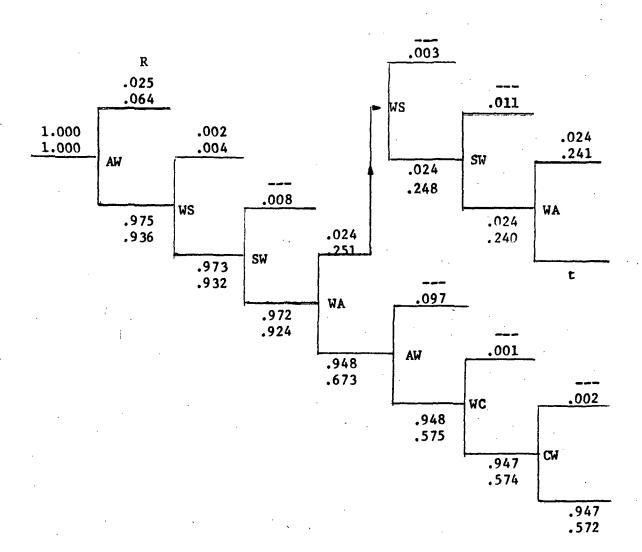
the Rayleigh type because the scattering phenomena is not strongly wavelength dependent. The model presented here can also be applied to the visible wavelengths if the appropriate complex indices of refraction of the leaf constituents in the visible wavelengths are known. The authors believe that the model of a leaf presented in this article is more complete and realistic than as proposed by Willstatter and Stoll. It supports the experimental results of Breece and Holmes, 17 and Wooley. 18

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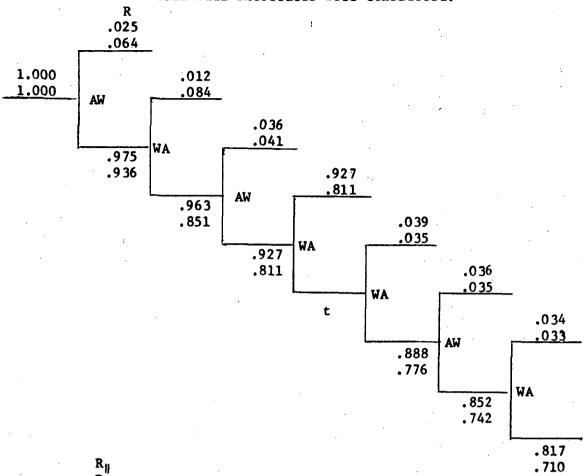
## Captions

- TABLE 1 (a) The values of the reflected and transmitted intensity of the ray at first seven interfaces. The optical mediums considered are cell wall, chloroplasts, cell sap and air.
- (b) The values of the reflected and transmitted intensity of the ray at first seven interfaces. Only cell wall air and air cell wall interfaces were considered. Figure 1. Pathway of light ray through the leaf cross section. R denotes the reflected ray. Solid lines show the pathway of light considering cell wall, chloroplasts, cell sap and air as the optical mediums. Dotted lines show the pathway of light considering only cell wall and air as the optical mediums. The numbers along the rays denote their total intensity. The rays whose total intensity is less than 0.018 are not shown. Figure 2. Pathway of light through a leaf as envisioned by the Willstatter and Stoll theory. (Taken from Sinclair<sup>4</sup>)

(a) The values of the reflected and transmitted intensity of the ray at first seven interfaces. The optical mediums considered are cell wall, chloroplasts, cell sap and air.



(b) The values of the reflected and transmitted intensity of the ray at first seven interfaces. Only cell wall - air and air - cell wall interfaces were considered.



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 $R_{||}$  = reflection || to the plane of incidence

 $R_{\parallel}$  = reflection  $\perp$  to the plane of incidence

 $T_{||}$  = transmission || to the plane of incidence

 $\underline{\mathbf{T}}_{\perp}$  = transmission  $\perp$  to the plane of incidence

-- denotes that the value of intensity is less than 0.0005

AW Air to Cell Wall

SW Cell Sap to Cell Wall

CW Chloroplasts to Cell Wall

SC Cell Sap to Chloroplasts

CS Chloroplasts to Cell Sap

WC Cell Wall to Chloroplasts

WS Cell Wall to Cell Sap

WA Cell Wall to Air

Note: Only those rays whose total intensity is more than 0.05 are shown in this table.

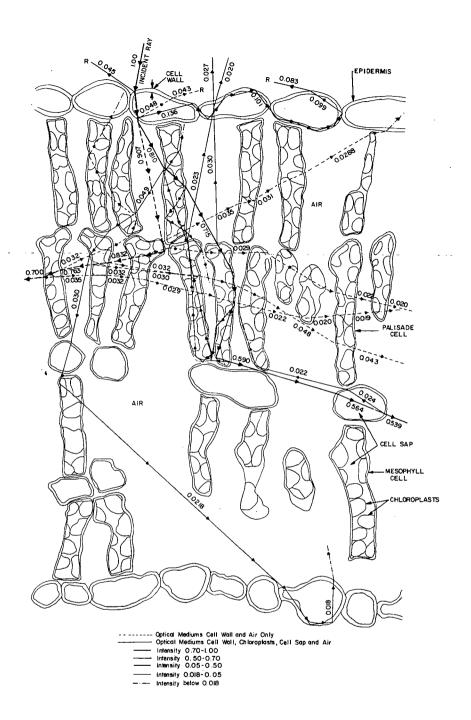


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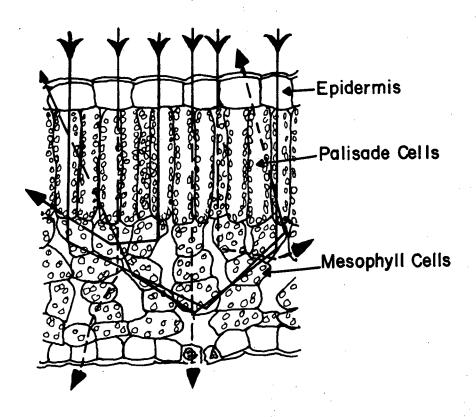


Figure 2. Pathway of light through a leaf as envisioned by Willstatter and Stoll theory.

(Taken from Sinclair4)

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